Application Note

Environmental

Agilent

Trusted Answers

A Method for PFAS Analysis in Drinking and Nonpotable Waters Using the Agilent 6495 LC/MS System

Direct aqueous injection method for the determination of 47 PFAS in water

Authors

David Powell and
Marcus Chadha
Agilent Technologies, Inc.

Laura Pinkney,
Beverley Kerrigan, and
Jenny Grimshaw
United Utilities PLC,
Warrington, UK

Abstract

A comprehensive liquid chromatography/triple quadrupole mass spectrometry (LC/MS/MS) method was developed and validated for the quantitation of 47 per- and polyfluoroalkyl substances (PFAS) with the intention to accelerate and simplify routine laboratory water testing. Compound transitions and optimized parameters were applied to the analytical method. The method suitability was demonstrated using an Agilent 1290 Infinity II LC system coupled to an Agilent 6495 triple quadrupole LC/MS on water, surface water, and ground water, using direct injection of samples.

Method performance was evaluated and validated according to ISO 17025 based on instrument limit of detection (LOD), limit of quantification (LOQ), calibration curve linearity, uncertainty of measurement (UoM) recovery, and precision using calibration standards up to 0.25 µg/L. All 47 of the analytes demonstrated linearity with R^2 ≥0.995. Method precision was assessed using relative standard deviation (RSD). RSD at 0.1 and 0.02 µg/L for all compounds were within the limit of 12.5% and mean recoveries were within the limits of 75 to 125% for all target analytes; UoM values were <60% for all compounds.

Introduction

PFAS are compounds of unique chemistry, with surfactant properties. As with many surfactants, PFAS often have a hydrophilic moiety covalently linked to a hydrophobic alkyl chain. In PFAS, the alkyl chain generally has few or zero hydrogen atoms; instead, the alkyl chain is populated with fluorine and other halogens. The diversity of the chain length, branching, and type of hydrophilic moieties are still being discovered today. However, regulatory bodies in many countries require monitoring of specific PFAS, thought to be end-products of degradation processes, in potable drinking water samples.

An LC/MS/MS method has been developed for the analysis of 47 PFAS compounds. This method, including sample preparation, chromatographic separation, and MS detection targets quantitation and results interpretation, helps streamline routine PFAS analysis, and therefore accelerates lab throughput and productivity. Details of sample preparation procedures and instrumentation setup will be discussed in conjunction with the data analysis parameters enabling the quantification and confirmation of PFAS in water by direct injection.

Experimental

Chemicals and reagents

LC/MS-grade solvents and analytical reagents were used for this study.

Standards and solutions

Ready-to-use and custom-premixed and individual PFAS standards were acquired where available. Neat compounds were sourced where custom mixes were not available.

Two intermediate standard mixes were prepared from stock standards and used for the rest of the experiments. Working standards were diluted from mixes 1 and 2 and used for the preparation of prespiked calibration and QC samples.

A separate internal standard mixture (IS mix) containing 23 stable isotope-labeled compounds was prepared from a custom mix.

Calibration standards were prepared in water. Serial dilutions were performed to prepare six calibration concentration levels. Calibration standards were freshly prepared and stored in a refrigerator at 3 °C, if not used immediately.

47 PFAS compounds

Analyte	CAS No.
PFBA	375-22-4
PFPeA	2706-90-3
PFHxA	307-24-4
PFHpA	375-85-9
PFOA	335-67-1
PFNA	375-95-1
PFDA	335-76-2
PFUnDA	2058-94-8
PFDoDA	307-55-1
PFTrDA	72629-94-8
PFBS	375-73-5
PFPeS	2706-91-4
PFHxS	355-46-4
PFHpS	375-92-8
PFOS	1763-23-1
PFNS	98789-57-2

Analyte	CAS No.
PFDS	335-77-3
PFUnS	749786-16-1
PFDoS	79780-39-5
PFTeDA (PFTeA)	376-06-7
PFHxDA	67905-19-5
PFODA	16517-11-6
HFPO-DA (Gen X)	62037-80-3
ADONA	958445-44-8
HFPO-TA	13252-14-7
PFMOPrA	377-73-1
NFDHA	151772-58-6
PFMOBA	863090-89-5
PFecHS	133201-07-7
3:3 FTCA	356-02-5
5:3 FTCA	914637-49-3
7:3 FTCA	812-70-4

Analyte	CAS No.
PFEESA	113507-82-7
6:2 Cl-PFESA; 9Cl-PF3ONS	756426-58-1
8:2 Cl-PFESA; 11Cl-PF3OUdS	763051-92-9
4:2 FTSA; 4:2 FTS	757124-72-4
6:2 FTSA; 6:2 FTS.	27619-97-2
8:2 FTSA; 8:2 FTS	39108-34-4
FBSA	30334-69-1
FHxSA	41997-13-1
FOSA	754-91-6
MeFOSA; N-MeFOSA	31506-32-8
EtFOSA; N-EtFOSA	4151-50-2
MeFOSE	24448-09-7
EtFOSE	1691-99-2
NMeFOSAA; MeFOSAA	2355-31-9
NEtFOSAA; EtFOSAA	2991-50-6

Sample preparation

PFAS-free equipment was used throughout the sample preparation and all equipment was tested for the 47 compounds before use to ensure that PFAS were not present, in order to prevent contamination of samples.

Once the addition of reagents to the samples was complete, the samples were transferred to analytical vials and analyzed by direct injection.

Instrumentation

Chromatographic separation was performed using an Agilent ZORBAX Rapid Resolution High Definition Eclipse Plus C18 column installed on a 1290 Infinity II LC system.

The individual modules of the 1290 Infinity II LC system included:

- Agilent 1290 Infinity II binary pump (G7120A)
- Agilent 1290 Infinity II multisampler (G7167B)
- Agilent 1290 Infinity II multicolumn thermostat column compartment (G7116B)

An Agilent 6495 liquid chromatography/triple quadrupole mass spectrometer (G6495C) with an Agilent Jet Stream (AJS) electrospray ion source was operated in dynamic multiple reaction monitoring (dMRM) mode. This allows the addition of more MRM transitions if future development is required for additional compounds. The LC/TQ autotune was performed in unit and wide modes. All data acquisition and processing were performed using the Agilent MassHunter software (version 10).

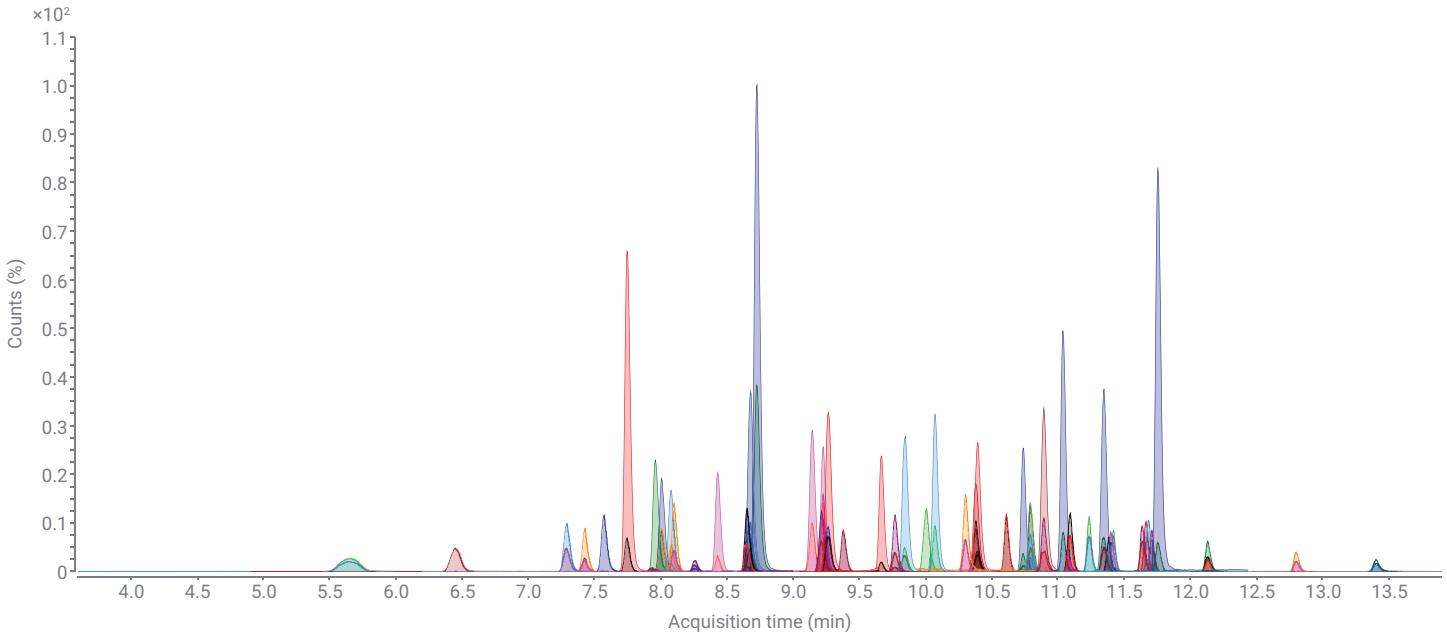
Results and discussion

A major part of this work was the development of dynamic MRM transitions for all 47 PFAS compounds. For each compound, the MRM transitions, as well as collision energies and ionization polarity, were optimized using Agilent MassHunter Optimizer software. The two most abundant product ions per compound were selected automatically, where available. Depending on the fragmentation behavior of the individual compound, two MRM transitions were selected per compound (except where only one transition was stable enough to be monitored). This selection was done to give confidence for identification and confirmation by LC/MS/MS. The two most abundant fragments were defined as primary transitions that were acquired over the retention time window, then used as the quantifier and qualifier ion.

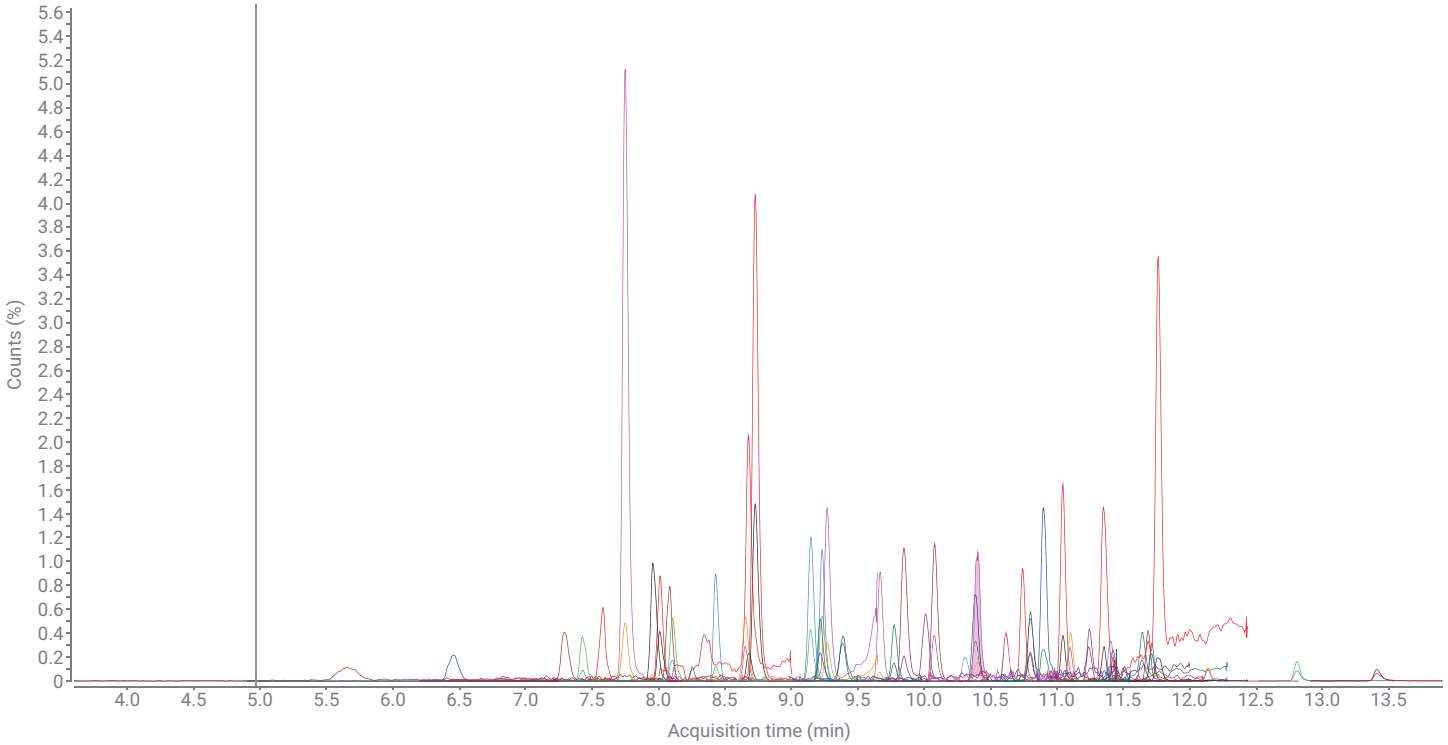
The chromatographic method was optimized using the ZORBAX RRHD Eclipse Plus C18 column, which resulted in good separation and distribution of 47 PFAS within an 18-minute HPLC gradient. The flow rate offered effective desolvation of target ions using the AJS ion source.

The selected cycle time ensured that sufficient data points were collected across the chromatographic peaks for reproducible quantitation and conformation of results.

250 ng/L standard



10 ng/L standard



Verification of method performance and validation

The method performance criteria were assessed, and validation was completed based on linearity, method sensitivity, UoM, recovery, and precision. The validation was completed using twelve 11 × 2 batches containing blanks and spiked standards and four real matrix samples spiked at blank, low, and high levels. Water matrices used included drinking water, surface water, and ground water. All compounds met the requirements of the validation process for precision (<12.5%), bias (<25%), UoM (<60%), limits of detection (<0.01 µg/L), and limits of quantification (<0.03 µg/L). This method is accredited by United Kingdom Accreditation Service (UKAS) to meet the requirements of ISO 17025.

Conclusion

This application note describes a highly sensitive and reproducible method for the fast and reliable quantitation of 47 PFAS in water by direct injection. The dMRM method was created and optimized using MassHunter software and allows the addition of more MRM transitions if future development is required for additional compounds.

An Agilent 1290 Infinity II LC system coupled to an Agilent 6495 triple quadrupole LC/MS was used for the analysis. The 18-minute LC gradient method using an Agilent ZORBAX Rapid Resolution High Definition Eclipse Plus C18 column offered good chromatographic separation and even retention time distribution of all targets. LC/MS data acquisition was performed in dMRM mode with fast polarity switching for the most efficient use of instrument cycle time. The method performance was verified based on requirements for calibration curve linearity, instrument LOD, recovery, and precision. The results demonstrate the ability of the quantitative analytical method for 47 PFAS in water by direct injection.

www.agilent.com

DE42959526

Printed in the USA, March 10, 2023
5994-5887EN

Agilent
Trusted Answers